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[Continued on next page]

(54) Title: NOVEL PROTEINS AND NUCLEIC ACIDS ENCODING SAME

(57) Abstract: Disclosed herein are nucleic acid sequences that encode G-coupled protein-receptor related polypeptides. Also disclosed are polypeptides encoded by these nucleic acid sequences, and antibodies, which immunospecifically-bind to the polypeptide, as well as derivatives, variants, mutants, or fragments of the aforementioned polypeptide, polynucleotide, or antibody. The invention further discloses therapeutic, diagnostic and research methods for diagnosis, treatment, and prevention of disorders involving any one of these novel human nucleic acids and proteins.

WO 01/74904 A2



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**WHAT IS CLAIMED IS:**

1. An isolated polypeptide comprising an amino acid sequence selected from the group consisting of:
  - (a) a mature form of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83;
  - (b) a variant of a mature form of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein one or more amino acid residues in said variant differs from the amino acid sequence of said mature form, provided that said variant differs in no more than 15% of the amino acid residues from the amino acid sequence of said mature form;
  - (c) an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83; and
  - (d) a variant of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein one or more amino acid residues in said variant differs from the amino acid sequence of said mature form, provided that said variant differs in no more than 15% of amino acid residues from said amino acid sequence.
2. The polypeptide of claim 1, wherein said polypeptide comprises the amino acid sequence of a naturally-occurring allelic variant of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83.
3. The polypeptide of claim 2, wherein said allelic variant comprises an amino acid sequence that is the translation of a nucleic acid sequence differing by a single nucleotide from a nucleic acid sequence selected from the group consisting of SEQ ID NOS:1, 3, 4, 6, 8, 10, 12, 14, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, 39, 41, 80, and 82.

4. The polypeptide of claim 1, wherein the amino acid sequence of said variant comprises a conservative amino acid substitution.
5. An isolated nucleic acid molecule comprising a nucleic acid sequence encoding a polypeptide comprising an amino acid sequence selected from the group consisting of:
  - (a) a mature form of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83;
  - (b) a variant of a mature form of an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein one or more amino acid residues in said variant differs from the amino acid sequence of said mature form, provided that said variant differs in no more than 15% of the amino acid residues from the amino acid sequence of said mature form;
  - (c) an amino acid sequence selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83;
  - (d) a variant of an amino acid sequence selected from the group consisting SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein one or more amino acid residues in said variant differs from the amino acid sequence of said mature form, provided that said variant differs in no more than 15% of amino acid residues from said amino acid sequence;
  - (e) a nucleic acid fragment encoding at least a portion of a polypeptide comprising an amino acid sequence chosen from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, or a variant of said polypeptide, wherein one or more amino acid residues in said variant differs from the amino acid sequence of said mature form, provided that said variant differs in no more than 15% of amino acid residues from said amino acid sequence; and
  - (f) a nucleic acid molecule comprising the complement of (a), (b), (c), (d) or (e).
6. The nucleic acid molecule of claim 5, wherein the nucleic acid molecule comprises the nucleotide sequence of a naturally-occurring allelic nucleic acid variant.

7. The nucleic acid molecule of claim 5, wherein the nucleic acid molecule encodes a polypeptide comprising the amino acid sequence of a naturally-occurring polypeptide variant.
8. The nucleic acid molecule of claim 5, wherein the nucleic acid molecule differs by a single nucleotide from a nucleic acid sequence selected from the group consisting of SEQ ID NOS:1, 3, 4, 6, 8, 10, 12, 14, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, 39, 41, 80, and 82.
9. The nucleic acid molecule of claim 5, wherein said nucleic acid molecule comprises a nucleotide sequence selected from the group consisting of:
  - (a) a nucleotide sequence selected from the group consisting of SEQ ID NOS:1, 3, 4, 6, 8, 10, 12, 14, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, 39, 41, 80, and 82;
  - (b) a nucleotide sequence differing by one or more nucleotides from a nucleotide sequence selected from the group consisting of SEQ ID NOS:1, 3, 4, 6, 8, 10, 12, 14, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, 39, 41, 80, and 82, provided that no more than 20% of the nucleotides differ from said nucleotide sequence;
  - (c) a nucleic acid fragment of (a); and
  - (d) a nucleic acid fragment of (b).
10. The nucleic acid molecule of claim 5, wherein said nucleic acid molecule hybridizes under stringent conditions to a nucleotide sequence chosen from the group consisting of SEQ ID NOS:1, 3, 4, 6, 8, 10, 12, 14, 15, 17, 19, 21, 23, 25, 27, 29, 31, 33, 35, 37, 39, 41, 80, and 82, or a complement of said nucleotide sequence.
11. The nucleic acid molecule of claim 5, wherein the nucleic acid molecule comprises a nucleotide sequence selected from the group consisting of:
  - (a) a first nucleotide sequence comprising a coding sequence differing by one or more nucleotide sequences from a coding sequence encoding said amino acid sequence, provided that no more than 20% of the nucleotides in the coding sequence in said first nucleotide sequence differ from said coding sequence;

- (b) an isolated second polynucleotide that is a complement of the first polynucleotide; and
  - (c) a nucleic acid fragment of (a) or (b).
- 12. A vector comprising the nucleic acid molecule of claim 11.
- 13. The vector of claim 12, further comprising a promoter operably-linked to said nucleic acid molecule.
- 14. A cell comprising the vector of claim 12.
- 15. An antibody that binds immunospecifically to the polypeptide of claim 1.
- 16. The antibody of claim 15, wherein said antibody is a monoclonal antibody.
- 17. The antibody of claim 15, wherein the antibody is a humanized antibody.
- 18. A method for determining the presence or amount of the polypeptide of claim 1 in a sample, the method comprising:
  - (a) providing the sample;
  - (b) contacting the sample with an antibody that binds immunospecifically to the polypeptide; and
  - (c) determining the presence or amount of antibody bound to said polypeptide, thereby determining the presence or amount of polypeptide in said sample.
- 19. A method for determining the presence or amount of the nucleic acid molecule of claim 5 in a sample, the method comprising:
  - (a) providing the sample;
  - (b) contacting the sample with a probe that binds to said nucleic acid molecule; and
  - (c) determining the presence or amount of the probe bound to said nucleic acid molecule,thereby determining the presence or amount of the nucleic acid molecule in said sample.

20. The method of claim 19 wherein presence or amount of the nucleic acid molecule is used as a marker for cell or tissue type.
21. The method of claim 20 wherein the cell or tissue type is cancerous.
22. A method of identifying an agent that binds to a polypeptide of claim 1, the method comprising:
  - (a) contacting said polypeptide with said agent; and
  - (b) determining whether said agent binds to said polypeptide.
23. The method of claim 22 wherein the agent is a cellular receptor or a downstream effector.
24. A method for identifying an agent that modulates the expression or activity of the polypeptide of claim 1, the method comprising:
  - (a) providing a cell expressing said polypeptide;
  - (b) contacting the cell with said agent, and
  - (c) determining whether the agent modulates expression or activity of said polypeptide,whereby an alteration in expression or activity of said peptide indicates said agent modulates expression or activity of said polypeptide.
25. A method for modulating the activity of the polypeptide of claim 1, the method comprising contacting a cell sample expressing the polypeptide of said claim with a compound that binds to said polypeptide in an amount sufficient to modulate the activity of the polypeptide.
26. A method of treating or preventing a GPCR<sub>X</sub>-associated disorder, said method comprising administering to a subject in which such treatment or prevention is desired the polypeptide of claim 1 in an amount sufficient to treat or prevent said GPCR<sub>X</sub>-associated disorder in said subject.
27. The method of claim 26 wherein the disorder is selected from the group consisting of cardiomyopathy and atherosclerosis.

28. The method of claim 26 wherein the disorder is related to cell signal processing and metabolic pathway modulation.
29. The method of claim 26, wherein said subject is a human.
30. A method of treating or preventing a GPCR<sub>X</sub>-associated disorder, said method comprising administering to a subject in which such treatment or prevention is desired the nucleic acid of claim 5 in an amount sufficient to treat or prevent said GPCR<sub>X</sub>-associated disorder in said subject.
31. The method of claim 30 wherein the disorder is selected from the group consisting of cardiomyopathy and atherosclerosis.
32. The method of claim 30 wherein the disorder is related to cell signal processing and metabolic pathway modulation.
33. The method of claim 30, wherein said subject is a human.
34. A method of treating or preventing a GPCR<sub>X</sub>-associated disorder, said method comprising administering to a subject in which such treatment or prevention is desired the antibody of claim 15 in an amount sufficient to treat or prevent said GPCR<sub>X</sub>-associated disorder in said subject.
35. The method of claim 34 wherein the disorder is diabetes.
36. The method of claim 34 wherein the disorder is related to cell signal processing and metabolic pathway modulation.
37. The method of claim 34, wherein the subject is a human.
38. A pharmaceutical composition comprising the polypeptide of claim 1 and a pharmaceutically-acceptable carrier.



39. A pharmaceutical composition comprising the nucleic acid molecule of claim 5 and a pharmaceutically-acceptable carrier.
40. A pharmaceutical composition comprising the antibody of claim 15 and a pharmaceutically-acceptable carrier.
41. A kit comprising in one or more containers, the pharmaceutical composition of claim 38.
42. A kit comprising in one or more containers, the pharmaceutical composition of claim 39.
43. A kit comprising in one or more containers, the pharmaceutical composition of claim 40.
44. A method for determining the presence of or predisposition to a disease associated with altered levels of the polypeptide of claim 1 in a first mammalian subject, the method comprising:
- (a) measuring the level of expression of the polypeptide in a sample from the first mammalian subject; and
  - (b) comparing the amount of said polypeptide in the sample of step (a) to the amount of the polypeptide present in a control sample from a second mammalian subject known not to have, or not to be predisposed to, said disease;
- wherein an alteration in the expression level of the polypeptide in the first subject as compared to the control sample indicates the presence of or predisposition to said disease.
45. The method of claim 44 wherein the predisposition is to cancers.

46. A method for determining the presence of or predisposition to a disease associated with altered levels of the nucleic acid molecule of claim 5 in a first mammalian subject, the method comprising:
- (a) measuring the amount of the nucleic acid in a sample from the first mammalian subject; and
  - (b) comparing the amount of said nucleic acid in the sample of step (a) to the amount of the nucleic acid present in a control sample from a second mammalian subject known not to have or not be predisposed to, the disease;
- wherein an alteration in the level of the nucleic acid in the first subject as compared to the control sample indicates the presence of or predisposition to the disease.
47. The method of claim 46 wherein the predisposition is to a cancer.
48. A method of treating a pathological state in a mammal, the method comprising administering to the mammal a polypeptide in an amount that is sufficient to alleviate the pathological state, wherein the polypeptide is a polypeptide having an amino acid sequence at least 95% identical to a polypeptide comprising an amino acid sequence of at least one of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, or a biologically active fragment thereof.
49. A method of treating a pathological state in a mammal, the method comprising administering to the mammal the antibody of claim 15 in an amount sufficient to alleviate the pathological state.
50. A method for the screening of a candidate substance interacting with an olfactory receptor polypeptide selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, or fragments or variants thereof, comprises the following steps:

- a) providing a polypeptide selected from the group consisting of the sequences of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, or a peptide fragment or a variant thereof;
  - b) obtaining a candidate substance;
  - c) bringing into contact said polypeptide with said candidate substance; and
  - d) detecting the complexes formed between said polypeptide and said candidate substance.
51. A method for the screening of ligand molecules interacting with an olfactory receptor polypeptide selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein said method comprises:
- a) providing a recombinant eukaryotic host cell containing a nucleic acid encoding a polypeptide selected from the group consisting of the polypeptides comprising the amino acid sequences SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83;
  - b) preparing membrane extracts of said recombinant eukaryotic host cell;
  - c) bringing into contact the membrane extracts prepared at step b) with a selected ligand molecule; and
  - d) detecting the production level of second messengers metabolites.
52. A method for the screening of ligand molecules interacting with an olfactory receptor polypeptide selected from the group consisting of SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83, wherein said method comprises:
- a) providing an adenovirus containing a nucleic acid encoding a polypeptide selected from the group consisting of polypeptides comprising the amino acid sequences SEQ ID NOS:2, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 81, and 83;
  - b) infecting an olfactory epithelium with said adenovirus;
  - c) bringing into contact the olfactory epithelium b) with a selected ligand molecule; and
  - d) detecting the increase of the response to said ligand molecule.



This information is presented graphically in the multiple sequence alignment given in Table 3E (with GPCR3 being shown on line 1) as a ClustalW analysis comparing GPCR3 with related protein sequences.

5

Table 3E. Information for the ClustalW proteins:

10

- 1) Novel GPCR3 (SEQ ID NO:9)
- 2) gi|9935442|ref|NP\_064688.1| odorant receptor S46 gene [Mus musculus] (SEQ ID NO:53)
- 3) gi|6532001|gb|AAD27596.2|AF121976.1 (AF121976) odorant receptor S19 [Mus musculus] (SEQ ID NO:54)
- 4) gi|11908211|gb|AAG41676.1 (AF137396) HOR 5Beta14 [Homo sapiens] (SEQ ID NO:55)
- 5) gi|9938014|ref|NP\_064686.1| odorant receptor S18 gene [Mus musculus] (SEQ ID NO:56)
- 6) gi|7305349|ref|NP\_038647.1| olfactory receptor 67 [Mus musculus] (SEQ ID NO:57)

	10	20	30	40	50	60
GPCR3	.....	.....	.....	.....	.....	.....
gi 9935442	.....	.....	.....	.....	.....	.....
gi 6532001	.....	.....	.....	.....	.....	.....
gi 11908211	.....	.....	.....	.....	.....	.....
gi 9938014	.....	.....	.....	.....	.....	.....
gi 7305349	.....	.....	.....	.....	.....	.....
	70	80	90	100	110	120
GPCR3	.....	.....	.....	.....	.....	.....
gi 9935442	.....	.....	.....	.....	.....	.....
gi 6532001	.....	.....	.....	.....	.....	.....
gi 11908211	.....	.....	.....	.....	.....	.....
gi 9938014	.....	.....	.....	.....	.....	.....
gi 7305349	.....	.....	.....	.....	.....	.....
	130	140	150	160	170	180
GPCR3	.....	.....	.....	.....	.....	.....
gi 9935442	.....	.....	.....	.....	.....	.....
gi 6532001	.....	.....	.....	.....	.....	.....
gi 11908211	.....	.....	.....	.....	.....	.....
gi 9938014	.....	.....	.....	.....	.....	.....
gi 7305349	.....	.....	.....	.....	.....	.....
	190	200	210	220	230	240
GPCR3	.....	.....	.....	.....	.....	.....
gi 9935442	.....	.....	.....	.....	.....	.....
gi 6532001	.....	.....	.....	.....	.....	.....
gi 11908211	.....	.....	.....	.....	.....	.....
gi 9938014	.....	.....	.....	.....	.....	.....
gi 7305349	.....	.....	.....	.....	.....	.....
	250	260	270	280	290	300
GPCR3	.....	.....	.....	.....	.....	.....
gi 9935442	.....	.....	.....	.....	.....	.....
gi 6532001	.....	.....	.....	.....	.....	.....
gi 11908211	.....	.....	.....	.....	.....	.....
gi 9938014	.....	.....	.....	.....	.....	.....
gi 7305349	.....	.....	.....	.....	.....	.....
	310	320	330	340		
GPCR3	.....	.....	.....	.....		
gi 9935442	.....	.....	.....	.....		
gi 6532001	.....	.....	.....	.....		
gi 11908211	.....	.....	.....	.....		
gi 9938014	.....	.....	.....	.....		
gi 7305349	.....	.....	.....	.....		

